

SCREENING OF SESBANIA FOR TOLERANCE TO ALUMINUM TOXICITY AND SYMBIOTIC EFFECTIVENESS WITH ACID TOLERANT RHIZOBIA STRAINS IN ACID SOIL IN WESTERN KENYA

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SUMMARY

Nitrogen fixation by leguminous trees such as sesbania (*Sesbania sesban*) in acid soils is limited by aluminium (Al) toxicity and phosphorus (P) deficiency. We screened 214 East African sesbania accessions for Al toxicity tolerance, P use efficiency and sesbania–rhizobia symbiosis. Aluminium toxicity tolerance or sensitivity was measured by the relative root elongation index. Highly Al tolerant and sensitive accessions were screened for P use efficiency. Highly P use efficient and Al sensitive accessions were assessed for symbiotic effectiveness with acid tolerant rhizobia. Eighty-eight per cent of the accessions were Al toxicity tolerant. High Al levels reduced shoot P content by 88% and total dry matter (TDM) by 83%. P addition increased shoot P content and TDM. Rhizobia inoculation increased nodulation by 28–82%, shoot N content by 28–45% and TDM by 15–34% in the low rhizobia density acid soil of Bumala, Kenya. P use efficient accessions had higher nodulation, shoot N content and TDM in the ranges 32–70, 20–52 and 22–36%, respectively, compared to sensitive genotypes. The combination of sesbania accession (SSUG10) and rhizobia strain ASs48 was superior in shoot N accumulation. Inoculation of P use efficient germplasm with acid tolerant rhizobia can improve N-rich biomass accumulation suitable for N replenishment in acid soils.

INTRODUCTION

At present, there is an emphasis on the study and use of woody legumes in cropping systems to sustain crop production in nutrient deficient soils in sub-Saharan Africa (SSA) (Kiptot, 2006). Sesbania (*Sesbania sesban*) is one of the major tree legumes; it grows naturally and is planted in a wide range of environments in the East African region of Kenya, Tanzania and Uganda. It grows quickly and rapidly accumulates nitrogen (N)-rich biomass suitable for soil fertility replenishment and also provides fuel wood, fodder and mulch (Kusekwa *et al.*, 1991). However, the growth of tree legumes and functioning of N₂-fixing symbiotic bacteria in tropical soils is limited by soil acidity, since both rhizobia and the host tree legumes are sensitive to soil acidity (Howieson and Ewing, 1986). High acidity is associated with aluminium (Al),

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iron (Fe) and manganese (Mn) toxicities in the soil solution with corresponding deficiencies of available phosphorus (P), molybdenum (Mo) and calcium (Ca) (Giller and Wilson, 1991). In SSA, woody legumes have poor establishment and growth due to Al toxicity, P deficiency and ineffective N₂ fixation (Almeida *et al.*, 1981; Kisinyo *et al.*, 2005). In legumes, P deficiency in acid soils results in reduction in growth and N₂ fixation (Sanginga *et al.*, 1995). Low soil P levels slow the growth rate of some strains and completely prevent the growth of others (Keyser and Munns, 1979). Plants tolerant to Al toxicity can absorb and translocate more P from the roots to shoots and therefore grow better under acid soil conditions than those that are sensitive (de Miranda and Rowell, 1988). About 13% (7.5 million ha) of Kenya's land area has acid soils, mostly in the western region which comprises the highlands west of the Rift Valley, Nyanza and Western provinces (Kanyanjua *et al.*, 2002). Therefore, soil acidity is a major problem in Kenya since most of the high potential agricultural land is found in Western and Rift Valley provinces.

To enhance growth and N₂ fixation of sesbania in acid soils, use of both Al toxicity tolerant and P use efficient germplasm and their inoculation with acid tolerant rhizobia strains is important. While some sesbania germplasm growing in Kenyan acid soils have been screened for tolerance to Al toxicity and P use efficiency (Gudu and Odago, 2003), this has not been done for germplasm growing in Tanzanian and Ugandan acid soils. Compatible local acid-tolerant rhizobia strains have been isolated and used to enhance N₂ fixation and growth in sesbania and calliandra (*Calliandra calothyrsus*) in Kenyan acid soils (Gudu and Odago, 2003; Muok, 1997).

Farmers in SSA rarely use farm inputs such as fertilizers and/or rhizobia inoculation to grow agro-forestry tree legumes. The objective of the present study was to: i) screen various sesbania accessions growing in East African soils for tolerance to Al toxicity and low soil P; and ii) investigate symbiotic effectiveness between Al toxicity tolerant and P use efficient sesbania germplasm and their compatibility with local acid tolerant rhizobia inocula for the greatest agronomic and ecological relevance.

MATERIALS AND METHODS

Seed collection

Seeds of 214 sesbania accessions were collected from nine sites with acid soils (pH-H₂O < 5.5) in East Africa. These were from: Kenya (Kuinet (00°36'N, 35°18'E), Bumala (00°01'N, 29° 93'E), Kavutiri (00°25'S, 37°30'E) and Gitumba (00°40'S, 36° 56'E)); from Uganda (Mbale (01°06'S, 34°17'E), Tororo (00°63'S, 37°19'E) and Kabale (01°25'S, 29°93'E); and from Tanzania (Lushoto (04°79'S, 38°26'E) and Sokoine University of Agriculture (06°82'S, 37°63'E)). Seeds were obtained from single trees spaced at least about 200 m apart. The number of trees selected at each site depended on seed availability at the time of collection.

Soil characterization

Nine soil samples were taken at random from the surface (0–15 cm depth) at the experimental field site at Bumala, Kenya, and thoroughly mixed together to make

a composite sample, and about 0.5 kg of this sample was put in a polythene bag for laboratory analysis. It was air-dried, passed through a 2-mm sieve and analysed for pH-H₂O, organic carbon (C), total N, available P, exchangeable bases and Al according to procedures outlined in Okalebo *et al.* (2002). At the same time a large surface soil sample was taken as a composite from randomly sampled cores in a 90-kg nylon bag for the greenhouse study.

Seed pre-treatment

Seeds were surface sterilized in 3.5% calcium hypochlorite solutions by immersion for six minutes and rinsed three times in sterile distilled water. To promote fast germination, seeds were then soaked in water at 90 °C for 12 h to soften the seed-coat and incubated at 27 °C for 60 h on moist paper towels in a tray.

Experiment 1: Screening for tolerance to Al toxicity in nutrient solution

Seeds were pre-germinated as described above. Sixty healthy seedlings per accession, with equal radicle length, were transferred to Styrofoam sheets at the rate of six seedlings per accession. Each seedling was placed in a 3-mm diameter hole, held in place with small Styrofoam balls, and sheets were floated in trays containing 8.0 l of distilled water aerated with a pump. After 24 h, initial seminal root length (ISRL) was measured using a ruler. Distilled water was then replaced with a nutrient solution modified from Mugwira and Haque (1993), with the following salt concentrations: NH₄NO₃ (3 mM N); Ca(NO₃)₂·4H₂O (1 mM Ca); K₂SO₄ (1 mM K); KH₂PO₄ (1 mM P); Mg₂SO₄·7H₂O (1 mM Mg); H₃BO₃ (46 µM B); MnSO₄ (9 µM Mn); ZnSO₄·7H₂O (0.7 µM Zn); CuSO₄·7H₂O (0.3 µM Cu); FeEDTA (75 µM Fe); and Na₂MoO₄·7H₂O (0.07 µM Mo). To each nutrient solution, three concentrations of Al (0, 111 and 222 µM) in the form of AlCl₃·6H₂O were separately added. The nutrient solution was replaced with a fresh one every seven days and the final seminal root length (FSRL) was measured after 21 days. A completely randomized design (CRD) with three replicates was adopted for this experiment. Nutrient solution pH was maintained at 4.0 ± 0.5 daily with 0.1 M KOH or 0.1 M HCl. Relative root elongation index (RRE) was calculated for each accession as:

$$\text{RRE} = \frac{\text{NSRL}_{\text{treatment}}}{\text{NSRL}_{\text{control}}} \times 100$$

where NSRL is the Net Final Seminal Root Length = FSRL – ISRL. Seedlings were classified as tolerant, moderately tolerant and sensitive, when the RRE values were: > 70, 69–50 and < 50%, respectively.

Experiment 2: Screening of Al toxicity tolerant and sensitive accessions for P use efficiency

Accessions tolerant and sensitive to Al toxicity were selected from experiment 1 and were screened for their P use efficiency. The tolerant accessions were: SSBSA004 (Busia Kenya site No. 4), SSUG3 (Uganda site No. 3), SSUG4 (Uganda site No. 4), SSUG5 (Uganda site No. 5), SSUG6 (Uganda site No. 6), SSUG8 (Uganda site No. 8)

and SSUG10 (Uganda site No. 10). The sensitive accessions were: SSBSA203 (Busia Kenya site No. 203), SSUG7 (Uganda site No. 7) and SSUG9 (Uganda site No. 9). A sand screening method for P use efficiency was adapted from Zhu *et al.* (2001). Sand in sisal bags was washed with running tap water for two continuous days, air dried and passed through a 5 mm sieve. One kg portions of sand were weighed and mixed with the following chemicals at the indicated weights: NH_4NO_3 (0.3 g N), $\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$ (0.444 g Ca), K_2SO_4 (0.174 g K), $\text{Mg}_2\text{SO}_4 \cdot 7\text{H}_2\text{O}$ (0.185 g Mg), H_3BO_3 (0.5 mg B), MnSO_4 (0.6 mg Mn), $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ (2.2 mg Zn), $\text{CoSO}_4 \cdot 7\text{H}_2\text{O}$ (0.4 mg Co), $\text{CuSO}_4 \cdot 7\text{H}_2\text{O}$ (2 mg Cu), FeEDTA (0.4 mg Fe), $\text{Na}_2\text{MoO}_4 \cdot 7\text{H}_2\text{O}$ (0.5 mg Mo). Two levels of Al (low, 0 g; high, 0.051 g (222 μM Al) as $\text{AlCl}_3 \cdot 6\text{H}_2\text{O}$ and three levels of P (low, 0 g; medium, 0.75 g; high, 2.0 g P) as KH_2PO_4 were added. The chemicals were mixed with sand individually for each pot and then put in a 1.0 l plastic container with a hole at the bottom. Containers were watered for one week before transplanting and reservoirs at the bottom of each container were used to tap the leachates, which were put back into the container every two days. A CRD with three replications was used in this experiment.

Four healthy seedlings, pre-germinated as described above, were planted, leaving only the cotyledon above the sand culture. They were thinned a week later to two per pot and grown for 30 days. At harvesting time, plants were placed on a sieve and roots were carefully washed under a gentle stream of tap water. Shoots were separated from roots by cutting at the root collar. Roots and shoots were separately chopped into about 2–5 cm pieces, oven dried at 70 °C for 72 hours and their dry weights recorded. Shoot P content was determined by tissue digestion and sample solution concentrations absorbance read at a wavelength of 880 nm in a colorimeter according to procedures outlined in Okalebo *et al.* (2002).

Experiment 3: Screening for acid tolerant legume-rhizobia symbiotic effectiveness

Based on the results from experiment 2, seeds from five sesbania accessions (SSBSA004, SSUG4, SSUG5, SSUG6 and SSUG10) both Al toxicity tolerant and P use efficient, and one Al sensitive and P use inefficient (SSUG7) accession were pre-germinated as described above. Acid soil high in Al and low in P, obtained from Bumala, Kenya, was air dried, ground and passed through a 5-mm sieve. Polythene bags were filled with 2 kg soil each after mixing with 0.027 g P (30 k P ha^{-1}) as triple super phosphate and watered to field capacity prior to planting. Acid tolerant *Rhizobia* isolates ACc1, ACc13, ACc20, ACc25, ASs31 and ASs48 isolated from East African soils were obtained from the Kenya Forestry Research Institute Headquarters, Muguga. The inoculum for each isolate was mixed with distilled water to make a slurry of which 1.0 ml was used to inoculate the root collars of the test accessions. Seedlings were grown in the greenhouse for three months (i.e. sown on 13 March and harvested on 13 May 2007). The average temperature and relative humidity during the growing period were 30–38 °C and 50–75 %, respectively. The experiment was a 6 \times 7 factorial in a CRD with three replicates. The test accessions were watered to field capacity throughout the growing period.

During harvesting, shoots from each treatment were cut at the root collar and chopped into 2–5 cm pieces, dried at 70 °C for 48 h and weighed. Shoot P and N contents were analysed according to procedures outlined in Okalebo *et al.* (2002). Roots were placed on a sieve and carefully washed under a gentle stream of tap water. Nodules were detached from the roots, numbers were recorded and checked for N₂ fixation effectiveness by splitting open samples from each plant. Red-pink nodules (the presence of leghaemoglobin) indicated effective nodulation while whitish or greenish nodule denoted ineffective nodulation. Roots were oven dried at 70 °C for 48 h and their dry weight recorded.

Statistical analysis

The data were subjected to analysis of variance (ANOVA) using the GenStat computer package (Buysse, 2006) and means were separated using least significant difference (*l.s.d.*) whenever treatment effects were significant at $p < 0.05$.

RESULTS

Soil chemical characteristics

Bumala soil is strongly acidic, high in Al saturation, moderate in organic carbon and low in available P and total N (Table 1).

Experiment 1: Screening for tolerance to Al toxicity in nutrient solution

Based on the RRE classification, 70% (150 accessions) were found to be tolerant, 18% (39 accessions) moderately tolerant and 12% (25 accessions) sensitive to Al toxicity.

Experiment 2: Screening of Al toxicity tolerant and susceptible accessions for P use efficiency

High Al concentration reduced shoot P content by 88%, root dry matter by 75%, shoot dry matter by 88% and total dry matter (TDM) by 83%. Phosphorus additions at the medium (0.75 g P kg⁻¹ sand) rate increased shoot P by 85%, root dry matter by 104%, shoot dry matter by 125% and TDM by 119%, while the high (2.0 g P kg⁻¹ sand) rate increased shoot P by 154%, root dry matter by 121%, shoot dry matter by 151% and TDM by 130% (Table 2).

Aluminium tolerant accessions had higher shoot P uptake and dry matter accumulation than the sensitive accessions under stressful Al conditions (Table 3). There was a linear relationship between shoot P and seedling TDM accumulation ($R^2 = 0.93; y = 0.287x + 0.138$). The most Al toxicity tolerant accession SSUG10 had the highest shoot P and dry matter accumulation followed by SSUG6, while the most sensitive accession SSBSA203 had the lowest shoot P and dry matter accumulation.

Experiment 3: Screening for acid tolerant legume-rhizobia symbiotic effectiveness

Inoculation with rhizobia increased nodulation by 31–82%, shoot N by 28–45% and TDM 15–34% compared to the control (Table 4). Legume–rhizobia combinations with the highest number of nodules did not necessarily have the highest shoot N and

Table 1. Some chemical characteristics of the Bumala soil.

pH (H ₂ O)	Olsen P (mg P kg ⁻¹)	C (%)	Total N (%)	Exchangeable cations (cmol kg ⁻¹)				ECEC (cmol kg ⁻¹)	Al saturation (%)
				K	Mg	Ca	Al		
5.0 ± 0.03	5.23 ± 0.02	1.66 ± 0.01	0.11 ± 0.01	0.4 ± 0.01	1.6 ± 0.03	3.2 ± 0.02	2.0 ± 0.03	7.2 ± 0.03	27.8 ± 0.39

Means based on three replicates.

ECEC: Effective Cation Exchange Capacity.

Table 2. Effect of aluminum toxicity and soil phosphorus on seedling shoot phosphorus and dry matter.

P/kg sand (g)	Total shoot P (mg /plant) Al (μ M)			Root dry matter (g/plant) Al (μ M)			Shoot dry matter (g) per plant Al (μ M)			Total dry matter (g/ plant) Al (μ M)		
	0	222	Mean	0	222	Mean	0	222	Mean	0	222	Mean
0	7.23 \pm 0.56	0.86 \pm 0.08	4.05	0.75 \pm 0.08	0.19 \pm 0.03	0.48	1.32 \pm 0.08	0.16 \pm 0.01	0.72	2.08 \pm 0.12	0.36 \pm 0.03	1.22
0.75	9.29 \pm 1.03	5.68 \pm 0.54	7.48	1.08 \pm 0.09	0.88 \pm 0.09	0.98	1.74 \pm 0.09	1.50 \pm 0.10	1.62	2.82 \pm 0.17	2.51 \pm 0.19	2.67
2.00	12.27 \pm 0.66	8.33 \pm 0.57	10.30	1.22 \pm 0.08	0.90 \pm 0.10	1.06	2.00 \pm 0.09	1.61 \pm 0.09	1.81	3.22 \pm 0.15	2.37 \pm 0.15	2.80
Mean	9.60	4.95		1.02	0.66		1.67	1.09		2.70	1.74	
<i>s.e.d.</i> Al		0.05			0.05			0.05			0.07	
<i>s.e.d.</i> P		0.56			0.06			0.06			0.09	
<i>s.e.d.</i> Al \times P		0.79			0.09			0.08			0.13	

Means based on three replicates.

Table 3. Mean sesbania seedling shoot phosphorus and dry matter in sand culture.

	Shoot P (mg)	Root dry matter (g)	Shoot dry matter (g)	Total dry matter (g)
SSBSA203 [†]	4.40 ± 0.68 _f	0.52 ± 0.09 _f	0.93 ± 0.12 _f	1.45 ± 0.18 _e
SSUG3	7.16 ± 0.75 _{bcd}	0.73 ± 0.10 _{edf}	1.23 ± 0.14 _e	1.81 ± 0.19 _d
SSUG7 [†]	5.61 ± 0.80 _{ef}	0.60 ± 0.09 _{ef}	1.19 ± 0.14 _e	1.83 ± 0.20 _d
SSUG9 [†]	5.83 ± 0.78 _{d_{ef}}	0.62 ± 0.09 _{d_{ef}}	1.18 ± 0.14 _e	1.91 ± 0.19 _d
SSBSA004	6.45 ± 0.82 _{cde}	0.72 ± 0.08 _{edf}	1.30 ± 0.16 _{de}	2.03 ± 0.22 _{cd}
SSUG4	7.76 ± 1.01 _{bcd}	0.85 ± 0.10 _{cd}	1.52 ± 0.17 _c	2.35 ± 0.27 _{bc}
SSUG5	7.90 ± 1.09 _{bc}	0.98 ± 0.11 _{bc}	1.48 ± 0.19 _c	2.46 ± 0.30 _b
SSUG8	8.03 ± 1.06 _{bc}	0.76 ± 0.09 _{d_e}	1.72 ± 0.20 _b	2.48 ± 0.28 _b
SSUG6	8.54 ± 1.18 _b	1.08 ± 0.15 _b	1.45 ± 0.16 _{cd}	2.53 ± 0.29 _b
SSUG10	11.07 ± 1.46 _a	1.55 ± 0.19 _a	1.88 ± 0.22 _a	3.43 ± 0.39 _a
<i>s.e.d.</i>	1.03	0.12	0.10	0.17

Means based on three replicates. Means with different subscript letters are significantly different.

[†]Al susceptible.

biomass accumulation. Therefore, high nodule number does not necessarily result in high N₂ fixation and biomass accumulation. Aluminium tolerant and P use efficient accessions had higher nodulation of 32–70%, shoot N of 20–52% and TDM of 22–36% over and above the sensitive accessions. Symbiotic effectiveness between Al tolerant sesbania accessions and acid tolerant rhizobia isolates were different as illustrated by the shoot N and biomass accumulations. Accession SSUG10–rhizobia strain Ass48 symbiosis was the most effective in N-rich biomass accumulation making it the most suitable for soil N replenishment for the acid soil in this study.

DISCUSSION

Acid soils, like that of Bumala, are common among the important agricultural soils of Kenya and also in many parts of SSA and are a constraint to crop production (Kanyanjua *et al.* 2002; London, 1991). In tropical soils, high leaching rates of both cations and anions due to high rainfall, parent materials of acidic origin and, in some cases, continuous application of acidifying chemical fertilizers such as diammonium phosphate or ammonium sulphate, are responsible for soil acidity (van Straaten, 2002; pp. 25–28). In acid soils, high levels of exchangeable Al³⁺ ions and corresponding deficiencies of P, Ca, Mg and K are the main constraints to crop production (Giller and Wilson, 1991; Landon, 1991). Soil acidity, and low P and N make these soils unable to sustain healthy plant growth and high crop yields (Okalebo *et al.*, 1997).

Most (88%) of the sesbania accessions from the areas sampled are tolerant to Al toxicity. Gudu and Odago (2003) found that most sesbania and calliandra growing naturally in Kenyan acid soils were tolerant to Al toxicity. Plants tolerant to Al toxicity normally have better root growth, water and essential nutrients absorption and biomass accumulation than sensitive varieties (de Miranda and Rowell, 1988; Kochian, 1995; Sierra *et al.*, 2003). P is one of the essential nutrients whose absorption is adversely affected by Al toxicity, particularly in sensitive plants.

Table 4. Symbiotic effect between sesbania accessions and rhizobia isolates on nodulation (a), (b) shoot N and biomass accumulation (c).

Accessions	Rhizobia isolates							Mean
	Control	ACc1	ACc13	ACc20	ACc25	ASs31	ASs48	
a) Number of root nodules per plant								
SSUG7 [†]	52 ± 6.69	78 ± 5.36	61 ± 4.04	77 ± 4.63	111 ± 8.81	119 ± 6.36	85 ± 6.60	83 _d
SSBSA004	78 ± 8.01	96 ± 8.76	96 ± 12.74	91 ± 9.87	162 ± 6.69	119 ± 17.95	126 ± 10.41	110 _{bc}
SSUG4	100 ± 5.24	121 ± 7.84	190 ± 13.86	103 ± 9.06	209 ± 10.73	162 ± 9.87	105 ± 10.73	141 _a
SSUG5	80 ± 3.93	121 ± 10.39	91 ± 4.36	106 ± 12.12	149 ± 8.66	168 ± 9.24	109 ± 8.74	118 _b
SSUG6	81 ± 11.57	104 ± 9.84	130 ± 13.91	128 ± 2.91	119 ± 5.67	150 ± 7.51	147 ± 11.85	123 _b
SSUG10	74 ± 4.67	87 ± 7.80	126 ± 5.81	119 ± 8.95	119 ± 12.44	120 ± 10.27	163 ± 12.41	115 _b
Mean	77 _d	101 _c	116 _b	104 _{bc}	145 _a	140 _a	122 _b	
<i>s.e.d.</i> Rhiz.	6							
<i>s.e.d.</i> Ses.	5							
<i>s.e.d.</i> Rhiz. × Ses.	13							
b) Shoot nitrogen in milligrams per plant								
SSUG7 [†]	115 ± 11.5	171.5 ± 12.7	152.3 ± 16.9	151.6 ± 19.6	167.8 ± 6.7	164.5 ± 9.9	178.2 ± 5.9	157.3 _d
SSBSA004	171.8 ± 11.0	209.4 ± 15.7	197.9 ± 17.1	235.2 ± 16.9	227.0 ± 16.7	211.8 ± 7.0	265.9 ± 8.7	217.0 _{ab}
SSUG4	177.9 ± 10.8	223.7 ± 15.3	212.3 ± 12.5	180.3 ± 9.9	265.4 ± 19.0	167.1 ± 9.76	226.6 ± 9.0	207.6 _{bc}
SSUG5	132.4 ± 6.1	200.3 ± 8.8	186.9 ± 8.0	189.0 ± 11.4	200.2 ± 10.8	144.8 ± 8.1	266.9 ± 10.6	188.6 _c
SSUG6	124.7 ± 8.8	184.5 ± 17.9	228.2 ± 10.5	198.3 ± 17.5	187.2 ± 9.2	207.3 ± 17.5	252.5 ± 7.9	197.5 _{bc}
SSUG10	173.1 ± 7.0	179.0 ± 14.0	287.0 ± 11.6	196.0 ± 12.0	257.0 ± 10.8	286.8 ± 11.6	292.7 ± 4.9	238.8 _a
Mean	149.2 _c	194.7 _b	210.8 _b	191.7 _b	217.4 _b	197.1 _b	247.1 _a	
<i>s.e.d.</i> Rhiz.	13.1							
<i>s.e.d.</i> Ses.	12.1							
<i>s.e.d.</i> Rhiz. × Ses.	32.1							
c) Total biomass in grams per plant								
SSUG7 [†]	7.54 ± 0.07	9.01 ± 0.18	8.94 ± 0.16	8.37 ± 0.30	9.16 ± 0.31	8.45 ± 0.21	9.44 ± 0.40	8.70 _c
SSBSA004	9.82 ± 0.62	11.44 ± 0.65	10.70 ± 0.53	11.17 ± 0.16	12.66 ± 0.46	10.91 ± 0.23	11.88 ± 0.37	11.23 _{ab}
SSUG4	9.04 ± 0.60	10.71 ± 0.36	11.22 ± 0.26	9.33 ± 0.63	13.97 ± 0.32	9.74 ± 0.20	10.80 ± 0.30	10.69 _b
SSUG5	8.06 ± 0.43	10.32 ± 0.53	12.62 ± 0.41	9.51 ± 0.30	10.55 ± 0.12	9.73 ± 0.42	13.67 ± 0.57	10.64 _b
SSUG6	7.89 ± 0.11	10.55 ± 0.36	11.88 ± 0.35	10.31 ± 0.41	10.59 ± 0.52	11.66 ± 0.53	11.90 ± 0.42	10.68 _b
SSUG10	9.15 ± 0.53	9.73 ± 0.24	13.91 ± 0.36	10.17 ± 0.14	12.23 ± 0.40	14.23 ± 0.39	13.08 ± 0.61	11.79 _a
Mean	8.58 _c	10.29 _{ed}	11.55 _{db}	9.81 _d	11.53 _{ab}	10.79 _{bc}	11.80 _a	
<i>s.e.d.</i> Rhiz.	0.21							
<i>s.e.d.</i> Ses.	0.20							
<i>s.e.d.</i> Rhiz. × Ses.	0.53							

Means based on three replicates. Means with different subscript letters are significantly different.

[†]Al susceptible.

P is essential for root growth and energy transfer processes in plants, so its deficiency affects general plant growth (Tisdale and Nelson, 1975). Therefore the observed growth increase due to its application in this study is unsurprising. Tolerance to Al toxicity and P use efficiency in plants observed is an important tool that can be used to select suitable sesbania accessions for acid soils prevalent in SSA.

Sesbania inoculation with rhizobia increased nodulation and seedling growth in Bumala soil. Normally, when the indigenous rhizobia populations are below 10^3 cells g^{-1} , inoculation is necessary for effective nodulation (Mulongoy and Ayanaba, 1986). Bumala soils have been reported to have low indigenous rhizobia populations (113 cells g^{-1} soil) compatible with sesbania (Odee, unpublished data). In a similar study, Makatiani and Odee (2007) reported increased nodulation, shoot N and sesbania growth after inoculation with superior rhizobia isolates in a Kenyan soil with a low indigenous rhizobia population. Similar differences in symbiotic effectiveness between sesbania accessions and acid tolerant rhizobia to those observed in Bumala have been reported elsewhere in Kenya (Muok, 1997). The accessions nodulated, fixed N_2 and accumulated biomass differently with different rhizobia isolates. The differences in symbiotic effectiveness between legume and rhizobia is an important tool that can be used to select superior combinations for N_2 fixation and faster growth suitable for soil N replenishment in acid soils.

CONCLUSIONS

Bumala soil is highly acidic, has low N, P and high Al. Most of the sesbania accessions naturally growing in acids soils of the areas sampled are tolerant to Al toxicity and among them there are P use efficient accessions. Inoculation of sesbania seedlings with acid tolerant rhizobia strains in acid soils with low indigenous rhizobia populations, such as in Bumala, can increase nodulation and N-rich biomass accumulation. It is apparent that Al toxicity, poor nodulation and/or ineffective N_2 fixation are a hindrance to sesbania growth in acid soils. Therefore inoculation of Al tolerant and P use efficient sesbania germplasm with superior acid tolerant rhizobia can enhance N-rich biomass accumulation, and is a suitable method for N replenishment in SSA acid soils.

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